

exotic equine infectious diseases and ii) assess existing infection control and quarantine measures on event horse yards. A questionnaire was designed, piloted and distributed electronically over six weeks. The target respondents were equestrians involved in eventing, including jumping their horses over fences either $\leq 100\text{cm}$ or $>100\text{cm}$. Data were described qualitatively and analysed statistically to identify any relationships between selected parameters. A total of 146 responses were analysed. The majority of respondents were female, involved as amateurs in eventing and their veterinary surgeon was the first choice for advice. To assess temperature, owners often used touch, but rectal temperature was taken rarely. The majority of horses were vaccinated against tetanus and influenza but a minority against EHV-1/-4. Most respondents identified the clinical signs of influenza and *Streptococcus equi*, but were less certain about EHV-1/-4. Only 30.1% of respondents had access to quarantine facilities, which were significantly more likely to be available in professional yards ($p=0.043$) and in riders competing above the 100cm level ($p=0.0003$). Professionals competing their horses above 100cm were significantly more likely to have quarantine facilities ($p<0.05$). A majority of yards with quarantine facilities isolated new horses for 2–4 weeks. Facilities included a separate stable (81.8%) or field (68.2%), but separate equipment (47.7%) and access to disinfectant (36.4%) were available less frequently. In conclusion, amateur equestrians involved in eventing require better education on the routine use of infection control and quarantine measures to minimise the impact of equine infectious diseases and thus strengthen infection control nationally.

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The use of PLFA analysis to detect differences in microbial activity in compost from horses treated with and without antibiotics

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There is increasing concern about the role of antibiotics and antibiotic resistant bacteria in the environment. In addition, there is an increasing recognition of the role of the gut microbiome in equine gastrointestinal health. Central to both of these concerns is the impact of oral antibiotics on horse health and incidence of pathogenic bacteria in equine feces. A study was therefore carried out to determine the impact of a common oral antibiotic administered to horses (Sulfadiazine-trimethoprim) on the microbial community of feces from horses receiving antibiotics. It was hypothesized that the microflora in equine stall waste collected from a herd of horses and composted in a static pile over 7 days would differ if these horses were treated with the prescribed antibiotic compared to horses not treated. A herd of 15 horses aged between 4 and 27 years received the recommended dose of a commercial oral antibiotic (12.5 g of sulfadiazine and 2.5 g trimethoprim per 1100 lb horse SID) for 4 days. Manure was collected from all horses from days 2 to 4 of antibiotic administration and composted in a static window approximately 4 feet high and 5 feet wide at the base. On days 0, 2, and 7, samples to fill a 1 gallon plastic bag were collected and stored at -20°C . For comparison, stall waste was also collected from the same horses for 3 days prior to antibiotic administration, composted in the same way, and sampled at 0, 2, and 7 days. Samples (approximately 10 g) were submitted at each time point from each pile for phospholipid fatty acid (PLFA) analysis. Results were then compiled and analyzed reporting the percentage of each class of microorganism according to fatty acid content. In samples from

horses not treated with these antibiotics, gram-positive fatty acids accounted for a larger percentage of the microbial community than gram-negative bacterial fatty acids. In samples from treated horses, both gram-positive and gram-negative bacterial fatty acid content stayed constant over time. This suggests the presence of Sulfadiazine-trimethoprim in compost inhibits the growth of gram-positive bacteria in comparison to gram-negative bacteria. While preliminary, these data suggest that administration of oral antibiotics to horses alters the microflora in composted equine stall waste.

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Elimination of the use of rubber stall mats for effective intervention of nosocomial *Salmonella enterica* spp. infection in a veterinary teaching hospital

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A nosocomial outbreak involving two serovars of *Salmonella enterica* spp. (serovars Javiana and Agona) in a population of hospitalized horses resulted in re-evaluation of current hospital biosecurity measures in the large animal veterinary teaching hospital (VTH). Evaluation of the VTH salmonella patient and environmental surveillance and tracking information was suggestive of certain trouble spots with repeated positive environmental and patient fecal samples. Interventional control measures included re-evaluating the standard operating biosecurity procedures, ensuring appropriate training sessions for hospital personnel, re-evaluating and modifying bedding and manure disposal, removing items considered to be non-cleanable, painting the interior of the facility with a nontoxic readily cleanable epoxy-type paint, replacement of stall walls with cleanable synthetic surfaces, and removing the non-cleanable rubber stall mats. These changes had previously been instituted in the equine and large animal intensive care unit except for the elimination of the rubber stall mats. When rubber stall mats were removed and replaced with a poured flooring material, the outbreak situation in the ICU was able to be controlled and subsequently, the main large animal hospital patient housing and care areas. Prevention of nosocomial infection requires a multifaceted approach and care must be taken to search out covert sources of contamination, such as the rubber stall mats, when standard intervention procedures do not prevent spread of the disease. Prevention of nosocomial infection is multi-modal and requires continued awareness, a committed team-oriented approach, and strict enforcement of biosecurity policies.

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Development of a high-throughput cell-based assay to determine anti-equine arteritis virus properties of chemical compounds

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Equine arteritis virus (EAV) is a single-stranded positive RNA virus that belongs to the *Arteriviridae* family, *Nidovirales* order. EAV can be transmitted either by respiratory or venereal route. Following the initial infection, stallions may become asymptomatic carriers and can shed the virus in their semen. Those stallions are reservoir for EVA and have to be properly handled to prevent any viral spread in horse population. Vaccination is an effective preventive measure to limit the spreading of the virus in equine population. However, no specific treatments to eliminate the virus from shedding stallions are currently available. The main objective of our study was to develop a high-throughput infection assay to screen candidate drugs for their antiviral properties against EAV. We have decided to use RK-13 cells since EAV replication is cytopathic in those cells. First, cytotoxic effects of different diluents (DMSO, Tween, PBS), likely to be used for compounds reconstitution, have been evaluated on RK-13 cells. In parallel, the same compounds were tested for their ability to prevent viral cytopathic effect in infected RK-13. Cell viability was evaluated using a commercial luciferase-based assay to quantify cellular ATP in culture wells. To determine the best model to test compounds, cells were cultured and infected in 96-well plates in the presence of EAV at different multiplicity of infection (MOI) and different incubation times (24h, 48h, 72h). Our results show that Tween 80 and PBS do not exhibit any toxicity on RK-13 cells and can be used as diluent to reconstitute compounds unlike DMSO that should be used at concentrations lower than 1%. Interestingly, Ribavirin, an inhibitor of inosine monophosphate (IMP) dehydrogenase leading in cells to purine (GTP pools) depletion, exhibits an anti-EAV activity at 20 µg/ml. In conclusion, we have developed a high-throughput cell-based assay using RK-13 cells that can be infected with EAV at different MOI. With this assay anti-EAV properties of compounds can be assessed in 48 hours. Compounds with a confirmed, *in-vitro*, anti-EAV action will be good candidates for *in-vivo* studies.

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Report of a case of Salmonellosis in an adult horse

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Salmonellosis is one of the main causes of chronic diarrhea in adult horses. Treatment is difficult and the prognosis is poor. In this work, a case of salmonellosis in an Argentinean polo race, castrated, 5 year-old horse, is reported. In July 2015 the horse arrives at a farm in Tandil District, in poor condition, with diarrhea and weakness. On August 24th, the veterinarian is consulted. The horse was sharing the farmyard with chickens; it was depressed, with edema of the four limbs and pale mucous membranes. It is treated with fenbendazole (10mg/kg/24 h for 5 days). For a better

follow up, on September 13th, it is moved to a pension in the city of Tandil. The horse had hyperthermia (40 °C), pale mucous membranes and profuse, watery diarrhea. It is treated with 1 mL/10-20 kg bodyweight/day of 20 g sulfadimidine, 4 g trimethoprim and 0.04 g loperamide-HCl for 3 days. Diet is changed to pellets, alfalfa and oats. Blood for blood-count is drawn on September 15th (Table 1). Parasitological examination of stool samples is negative. On September 30th, the horse is again depressed, with foreskin and ventral edema, hyperthermia (40.2 °C), pale mucous membranes, weakness and loss of appetite. Blood-count was performed (Table 1). Stools were collected for microbiological and parasitological tests. The coproparasitological results were negative.

Table 1

Alterations in the blood-count results

Parameter	Results 9/15	Results 9/30	Units
Hematocrit	28	14	%
Hemoglobin	9.5	9.2	g/dl
Erythrocytes	5.000.000	3.350.000	mm ³
M.C.V.	56	39	fl
M. C. H	19	14.1	Pg
Leukocytes	9600	16600	mm ³
Band neutrophils	0%	5	%
Basophils	2	0	%
Total proteins	7.3	7	g/dl
Albumin	2.3	2.18	g/dl
Globulins	5		g/dl
Bilirubin	2.5		mg%
Fibrinogen	400		mg%

On October 1st, hydration therapy and blood transfusion are performed. Penicillin 30,000 IU/kg every 24 h, 6.6 mg/kg/IV gentamicin every 12 h and flunixin meglumine at endotoxic doses for 5 days are administered. Stool samples were subject to the systematic processes for isolation and identification of microorganisms: stains, blood agar and McConkey cultures, pre-enrichment in selenite broth and subcultures in SS agar in aerophilic conditions at 37 °C. The 48 h subculture develops lactose negative colonies with sulfhydryl of a Gram negative bacillus, which is subject to TSI, LIA, catalase, oxidase, nitrates reduction, IMViC, urea and motility. These tests allowed the identification of *Salmonella* sp. The isolated strain was sensitive to: enrofloxacin, gentamicin, nalidixic acid, trimethoprim-sulfadiazine and ceftriaxone. On October 9th, the horse worsens, remains in recumbent position and it is decided to perform euthanasia. Microbial isolation, signs and blood biochemistry are in agreement with a case of chronic salmonellosis. Transmission by carriers or other co-habitant animals is possible. The determination of the species and serotype will allow further genetic studies.